REMARKS

Entry of the foregoing, reexamination and further and favorable reconsideration of the subject application in light of the following remarks, pursuant to and consistent with 37 C.F.R. § 1.112, are respectfully requested.

The Office Action Summary correctly indicates that claims 19-38 are pending in the application. Claims 19-38 are under consideration and stand rejected.

By the foregoing amendment, claims 19 and 35 have been amended. In particular, claim 19 has been amended to recite that the gel filtration step is carried out on a support comprising beads with a diameter of between 5 and 105 μm, to delete the word "approximately," and in the interest of concision to delete the unnecessary recitation of conventional elements of the gel filtration step. Support for the amendment to claim 19 can be found in the specification in at least page 15, line 4. Claim 35 has been amended to correct an obvious but inadvertent spelling error. No prohibited new matter has been introduced by way of the above amendments. Applicants reserve the right to file a continuation or divisional application on subject matter canceled by way of this Amendment.

Objection

Claim 35 was objected to for a misspelling. As discussed above, claim 35 has been corrected. Thus, withdrawal of this objection is respectfully requested.

Rejection under 35 U.S.C. § 103

The rejection of claims 19-38 under 35 U.S.C. § 103(a) as allegedly unpatentable over Shabram et al. WO 96/27677 A2 ("Shabram") in view of Berg WO 98/33572 A1 ("Berg") has been withdrawn. However, claims 19-38 have now been rejected under 35 U.S.C. § 103(a) as allegedly being unpatentable over Shabram in view of Berg and Bondoc et al., *J.*

Indust. Micro. & Biotech., 20:317-322, 1998 ("Bondoc"). The rejection as is respectfully traversed.

Applicants maintain that the rejection, even as modified in the outstanding Office Action, does not apply to the rejected claims, and applies even less to the claims as presently amended. As discussed in response to the previous Office Action, Shabram describes a method for purifying adenoviral particles comprising two chromatographic steps that consist in submitting a clarified cell lysate obtained from adenovirus infected cells to a DEAE anion exchange chromatography step (using a conventional gravity column), and the resulting DEAE eluate to an immobilized zinc affinity chromatography step (again on a conventional gravity column).

At page 9, lines 13-15, Shabram provides a generic disclosure with respect to chromatographic methods. It should be stressed that only one sentence in the whole document refers to fluidized bed chromatography. More tellingly, this sentence cites nearly every known techniques available in the art to perform chromatography, *i.e.* conventional packed bed (gravity), high pressure liquid chromatography using radial or axial flow, and batch and fluidized bed. Shabram merely discloses an all-inclusive listing that can be found in a number of basic books in connection with chromatography, and fails to render any specific method obvious. In addition, it has been asserted that Shabram discloses the use of cross-linked agarose column by referring to page 11, lines 27-28. It is respectfully pointed out that this paragraph of Shabram in fact refers to the use of a cross-linked agarose column for hydrophobic interaction chromatography which is not encompassed by the claimed method.

Thus, reading Shabram, the skilled person might appreciate that a variety of chromatographic techniques could be used in fluidized bed but would not be motivated to

make the choice of anion exchange chromatography in fluidized bed using particles of adsorbent comprising an agarose matrix and a central core comprising quartz, and dextran chains covalently coupled to the agarose matrix, on which are attached positively charged groups.

Furthermore, Shabram fails to suggest the combination of the two chromatographic steps recited in the claimed methods, *i.e.* an anion exchange chromatography in fluidized bed followed by a gel filtration chromatography. In fact, considering the work of Shabram and coworkers as a whole, one skilled in the art would not have expected the presently claimed method to have been effective. In this respect, the Examiner's attention is directed to Huyghe's article (*Human Gene Therapy*, 6:1403; 1995, previously submitted as Exhibit B of the preceding Amendment and Reply). This article originates from Shabram's team and can be viewed as a synthesis of the work described in the Shabram application cited by the Office. Shabram and coworkers investigated several types of chromatographic techniques for the purification of recombinant adenovirus particles. Specifically, anion exchange, gel filtration (*i.e.* size exclusion), hydrophobic interaction and metal chelating resins were tested. Dr Shabram and his co-workers reported that the recovery off the gel filtration column was very low with only 15-20% of the virus injected being eluted (see on page 1408, section "size exclusion chromatography"). Thus, upon consideration of all the evidence of record, Shabram cannot be considered to have directed one skilled in the art towards the presently claimed invention as proposed in the rejection. Berg and Bondoc fail to remedy the serious deficiencies of Shabram.

Berg reviews fluidized bed chromatographic techniques for separating a "substance" from a liquid sample using particles of adsorbent equipped with flexible extenders coated with a ligand having affinity to the substance. As illustrated in the section "Sample to be

applied," particles of adsorbent can be used for separation of a large variety of macromolecules, with molecular weight > 5,000 Daltons such as polysaccharides, proteins, polypeptides, nucleic acids and synthetic water-soluble polymers as well as compounds with molecular weight <5,000 Daltons (see on page 12 lines 7-13).

Berg does not suggest whatsoever that the fluidized-bed process could be effective for virus purification, and even less for adenovirus purification. It must be remembered that adenoviral particles are far more complex than the macromolecules considered by contrast as suitable for fluidized bed separation by Berg. Adenoviral particles are complex structures comprising a nucleic acid genome packaged in a capsid that consists of more than 10 polypeptides. The complex interactions between separate parts of the adenovirus must be preserved during the purification process in order to retain virus infectivity. Moreover, even if Berg states that there is no upper limit in molecular weight of compounds that can be separated by the fluidized bed chromatography, they nevertheless conclude that the fluidized-bed chromatography process *"is normally **limited** to the adsorption/separation of compounds that have **a molecular weight below 1,000,000 Daltons**"* (see on page 12 lines 13-16). Thus, while Applicants maintain that Berg teaches against the present invention, at the very least Berg teaches that it would have been considered unusual (and therefore not obvious) to utilize fluidized-bed chromatography for a compound substantially greater than $1x10^6$ Da. Adenoviral particles have a molecular weight of approximately $1.5x10^8$ Da, *i.e.* more than two orders of magnitude or about 150 fold above the normal limitation of the method as taught by Berg. Given the more than two orders of magnitude above the normal molecular weight limit given by Berg of the molecular weight of an adenoviral particle, according to Berg, it would be highly unusual and thus there would have been no reasonable expectation of success in using adsorbent particles of Berg to achieve a good purification profile and

retain most of the virus infectivity, let alone the surprising results that can be achieved using the presently claimed method.

Bondoc discloses a method of purifying recombinant adenoviral particles using gel filtration. Bondoc, alone or in combination with Shabram and Berg, fails to describe all the elements of the claimed method that combines an anion exchange chromatography in fluidized bed using specifically recited particles of adsorbent followed by a gel filtration chromatography carried out on a support comprising beads with a diameter of between 5 and 105μm and producing the surprising results as recited in the present claims.

In conclusion, all the elements of the presently claimed methods are neither described nor suggested in Shabram, Bondoc and Berg, either alone or in combination. Moreover, none of the cited references provides an incentive to the skilled person to combine the chromatographic steps comprising the claimed method for purifying adenoviral particles. There would have been no motivation provided by in Shabram, Berg, and/or Bondoc to choose a fluidized bed method out of the laundry list of other techniques Shabram. Mere knowledge of the existence of fluidized bed chromatographic techniques could not simply be translated into predictability of success, let alone the surprising effectiveness of the presently claimed methods, particularly when the work of Shabram and coworkers is considered as a whole, as evidenced by Huygue et al.

Considering that Dr. Shabram and his co-workers disclose that gel filtration leads to a very low viral yield (see Huygue et al., 1995, *supra*), and thus is not suited for the purification of recombinant adenoviral particles, there is no suggestion or motivation found anywhere in the art of record to modify Shabram's method by including the use of the adsorbent particles of Berg and the gel filtration chromatography step of Bondoc.

Bondoc contains no suggestion that would motivate one to combine the teachings of the references to arrive at the present invention. Furthermore, there was no way of knowing how effective the adsorbent particles of Berg would be for the purification of large-sized and complex macromolecules such as adenovirus. Given the 150 fold difference between the normal molecular weight limit given by Berg and the molecular weight of an adenoviral particle, a skilled person could not have expected an improvement of the total yields when using the adsorbent particles of Berg for the separation of adenoviral particles.

In light of the above, the methods of claims 19-38 are not obvious over the cited references separately or in combination. The prior art of record fails to teach all the elements of the claimed methods and fails to provide the required motivation to choose and combine even those elements that may be found mentioned in the prior art. Furthermore, it must be noted that claims 20-38 are directly or indirectly dependant on claim 19, and recite additional features that further distinguish those claims from the prior art of record. The allegations of the present rejection fail to show with specificity where each of the additional elements of claims 20-38 might allegedly be found in the prior art, and where in the prior art, the motivation to choose the recited combinations might allegedly be found. A properly supported *prima facie* case of obviousness has thus not been set forth in the outstanding Office Action.

For at least the foregoing reasons, the combination of references cited by the Examiner fail to render applicants' claimed invention obvious. Thus, withdrawal of the rejection as it applies to independent claim 19, as well as dependent claims 20-38, is proper and respectfully requested.

CONCLUSION

In view of the foregoing, further and favorable action in the form of a Notice of Allowance is believed to be next in order. Such action is earnestly solicited.

In the event that there are any questions relating to this application, it would be appreciated if the Examiner would telephone the undersigned concerning such questions so that prosecution of this application may be expedited.

Respectfully submitted,

BURNS, DOANE, SWECKER & MATHIS, L.L.P.

Date: October 13, 2004

By: _____
Christopher L. North
Registration No. 50,433

P.O. Box 1404
Alexandria, Virginia 22313-1404
(703) 836-6620